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STRUCTURAL AND FUNCTIONAL ORGANIZATION OF THE VESTIBULAR APPARATUS
IN RATS SUBJECTED TO WEIGHTLESSNESS FOR 19.5 DAYS ABOARD THE KOSMOS-
782 SATELLITE

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(NASA-TM-75529) STRUCTURAL AND FUNCTIONAL
ORGANIZATION OF THE VESTIBULAR APPARATUS IN
RATS SUBJECTED TO WEIGHTLESSNESS FOR 19.5
DAYS ABOARD THE KOSMOS-782 SATELLITE
(National Aeronautics and Space

N78-26717

Unclass

G3/51 23398

Translation of "Strukturnaya i funktsional'naya organizatsiya vestibul-
yarnogo apparata krysa, prebyvavshikh v usloviyakh nebesomosti
19.5 dney na sputnikye "Kosmos-782", Arkhiv Anatomii, Gistologii
i Embriologii, No. 1, 1978, pp. 22-28.



NATIONAL AERONAUTICS AND SPACE ADMINISTRATION
WASHINGTON, D. C. July, 1978

STANDARD TITLE PAGE

1. Report No. NASA TM 75229	2. Government Accession No.	3. Recipient's Catalog No.	
4. Title and Subtitle Structural and Functional Organization of the Vestibular Apparatus in Rats Subjected to Weightlessness for 19.5 days Aboard the Kosmos-782 Satellite		5. Report Date July, 1978	
		6. Performing Organization Code	
7. Author(s) Ya. A. Vinnikov, et al.		8. Performing Organization Report No.	
		10. Work Unit No.	
9. Performing Organization Name and Address SCITRAN Box 5456 Santa Barbara, CA 93108		11. Contract or Grant No. NASw-2791	
		13. Type of Report and Period Covered Translation	
12. Sponsoring Agency Name and Address National Aeronautics and Space Administration Washington, D.C. 20546		14. Sponsoring Agency Code	
15. Supplementary Notes Translation of "Strukturnaya i funktsional'naya organizatsiya vestibulyarnogo apparata krysa, prebyvavshikh v usloviyakh nebesomnosti 19.5 dney na sputnikye "Kosmos-782", Arkhiv Anatomii, Gistologii i Embriologii, No. 1, 1978, pp. 22-28			
16. Abstract The vestibular apparatus was investigated in rats subjected to weightlessness for 19.5 days aboard the Kosmos-782 satellite, including the effects of acceleration. Nineteen rats, nine of which served as controls, were studied. The vestibular apparatus was removed, and its sections were fixed in a glutaraldehyde [sic] solution for investigation by light and electron microscopes. Structural and functional changes were noted in the otolith portions of the ear, with the otolith particles clinging to the utricular receptor surface and with the peripheral arrangement of the nucleolus in the nuclei of the receptor cells. It is possible that increased edema of the vestibular tissue resulted in the destruction of some receptor cells and in changes in the form and structure of the otolith. In the horizontal crista, the cupula was separated. Six photographs are enclosed to illustrate these changes.			
17. Key Words (Selected by Author(s))		18. Distribution Statement Unclassified - Unlimited	
19. Security Classif. (of this report) Unclassified	20. Security Classif. (of this page) Unclassified	21. No. of Pages 11	22.

STRUCTURAL AND FUNCTIONAL ORGANIZATION OF THE VESTIBULAR APPARATUS /22*
IN RATS SUBJECTED TO WEIGHTLESSNESS FOR 19.5 DAYS ABOARD THE KOSMOS-
782 SATELLITE

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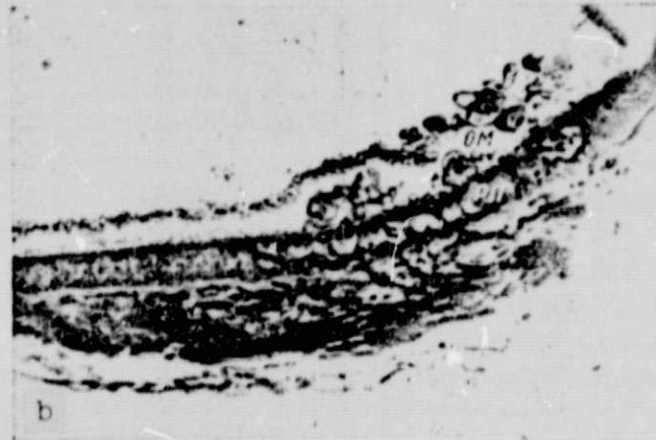
As has been proved by investigations of the vestibular apparatus, and in particular by researches on the utricles of the higher and lower vertebrates, acceleration causes a definite cycle of cytological displacements of the receptor cells which require a prolonged period for restoration (Ya. A. Vinnikov, et al., 1971; Ya. A. Vinnikov, 1971; Ya. A. Vinnikov, et al., 1972.) If these changes are imposed at the time of a space flight during weightlessness, then they may complicate the analysis of the effect on the vestibular apparatus. To eliminate this circumstance, research was conducted on the development of the vestibular apparatus on the embryos of fish and amphibians whose vestibular apparatus was still absent at the time of lift-off into space (Ya. A. Vinnikov and colleagues, 1972; Ya. A. Vinnikov, 1974; Ya. A. Vinnikov and colleagues, 1976). The results obtained showed that during their 40-hour stay under the conditions of space flight, 6 to 16 day old embryos of fish and amphibians continued their development. They displayed normal auditory vesicles, and then labyrinths with otoconiae in the case of the amphibians, and otoliths in the case of the fish. The cytological differentiation of the receptor cells was typical to a sufficient degree. The undesirable displacements caused by acceleration were absent. Nevertheless, on the 6th and 16th day of weightlessness some edema of

*Translator's note: Numbers in margin indicate pagination in foreign text.

the tissues was observed, which was more marked in comparison to the control, and in particular there was distortion of the correct crystalline form of the otoconiae in the amphibians and disorganization of the cyclicity of the daily otolith rings in the case of the fish. When analyzing the data regarding the vestibular apparatus of mammals (rats) subjected to weightlessness for 19.5 days on board the satellite "Kosmos-782", we attempted to differentiate between the changes which were related to acceleration (during lift-off and touchdown) on the one hand, and to weightlessness on the other hand. Special attention was given to the structure of the otoconiae and the otolith membrane.

Material and Method. All told, 19 rats were studied. 9 of them were control and 10 experimental. The vestibular apparatus was removed by the method of Ya. A. Vinnikov and L. K. Titova (1961). It was placed in a glutaraldehyde solution and its sections fixed for study, using /23 light and electron microscopes. Here, we take into account the period when the material was taken and how long it has remained in the fixative. It should be noted that in the case of all experimental rats, when the osseous labyrinth was dissected, hemorrhage was detected in the region of the base of the cochlea in the perilymph at the site of its transition into the vestibular perilymph.

Results Obtained. In the light microscope a well-preserved and properly-formed vestibular apparatus, otolithic membrane and otoliths were visible. The thinned-down otolithic membrane with the otoliths abutted on the surface of the receptor epithelium of the utricle. This was a consequence of the acceleration which the animal had undergone during the return of the spacecraft to the earth (Figure 1). Observations with both the light and electron microscopes revealed the presence of increased edema of the tissue and vacuolization of the cells (cf. Figure 2b), in comparison with the control (Figure 2a). In the connective tissue, stasis of the capillaries occurred. The cup-shaped and bud-shaped nerve endings around the type I and II receptor cells, as well as the synaptic structures, were sufficiently typical and were little different from the control. The nerve fibers and the supporting cells were especially well-supported. This is true of their microvilli, the organoids of the cytoplasm and the cell nuclei. The mitochondriae were, as a rule, compact, although they sometimes were swollen and vacuolated, especially in both the efferent and afferent endings. The receptor



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Figure 1: Utricle of the control rat (a); utricle of the rat subjected to the conditions of space flight for 19.5 days (b). Otolith membrane (OM) clings to the receptor surface (PN) of the utricle. Vertical section. Microphoto Ob. 7, oc. 10.

cells underwent somewhat more complicated changes. Obviously, in connection with the late periods of fixation the disappearance of the characteristic hernias from their apical surface was observed. They escape into the endolymph under the otolithic membrane in the form of "protruberances". These "protruberances" are coated with a prolongation of the plasmic membrane (cf. Figure 2b). They escape into the region of the rimae of the reticular membrane formed by the cuticle of the receptor cells and the desmosoma. In the cytoplasm of these protruberances individual swollen mitochondriae and even basal corpuscles of kinocillii were discovered, although, as a rule, the fascicle of stereocillii and the opposing kinocillii were well preserved. The cytoplasm of the type I and II receptor cells were characterized by increased charge density. The mitochondrial apparatus in the receptor cells was well preserved. The remaining cytoplasmic organelles were indistinguishable



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Figure 2: General view of the receptor epithelium in the utricle of the control rat and of the rat subjected to the conditions of space flight for 19.5 days. (a, b)

PK- receptor cell CT- stereocilia π - Protoplasmic protuberances HO- neuronal terminal. Electronogram.
Magnification: a- 2,900 b- 2,200

from the control. The nuclei of the receptor cells displayed a marginal distribution of the nucleoli; however, the latter did not escape beyond the limits of the nuclear membrane. Such a distribution of the nucleoli is usually the consequence of accelerations (Ya. A. Vinnikov and colleagues, 1971).

Together with the small changes in the receptor cells which have been described, individual cells, principally in Type I, were discovered with distinct symptoms of destruction and even of necrobiosis. As a rule, such cells were located inside the cup-shaped nerve endings, which

in turn were extremely vacuolized (Figure 3).

The otolithic membrane, which in the control has a granular structure, acquires a more diffuse, gelatinous character in the experimental animals. It is possible to trace the penetration of the kinocilii and the long stereocilii of the receptor cells into the otolithic membrane. They are slightly bent under the influence of the contiguity of the otolithic membrane to the receptor surface. The otoconiae embedded in it, which in the control have the proper form of polygonal crystals with a homogeneous alternating bright and dark granular composition (Figure 4a) are drastically changed in the experimental animals. Under the conditions of weightlessness, the otoconiae assume, as a rule, an oval-rounded form (cf. Figure 4b). Here the distribution of bright and dark substances constituting the otoconia is markedly altered. Thus, the center of the otoconia is a bright substance, and its periphery is filled with a dark substance, which takes on the character of distinct granules lying closely or loosely against each other.

Investigations of the saccules in the vestibular apparatus in these very same experimental animals have disclosed displacements of the structural organization - which in general is similar to the utricle - both in the light and electron microscope. In the crista, which proved to be more resistant, separation of the base of the cupula from the receptor epithelium occurred nevertheless, e.g., in the horizontal semicircular canal (Figure 5). However, the receptor cells and the innervation were practically unaffected in comparison to the control (Figure 6), although in connection with the separation of the cupula, the stereocilii were bent at their base. In this regard, further careful investigations are necessary. It is necessary to clarify the extent to which the changes in the structural and functional organization of the vestibular apparatus are reversible.

Summary. Data discussed show that the 19.5 day stay of the animals in conditions of weightlessness and two-fold acceleration has turned out to be somewhat variable as regards the structure and functional organization of the vestibular apparatus. The intensified function of the vestibular apparatus during accelerations alternated with its disconnection or modification under the conditions of weightlessness (Bracchi et al, 1975). Obviously, it is precisely this circumstance which gives rise to the result that (along with the considerable edema of the tissues



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Figure 3: Utricle of the rat subjected to the conditions of space flight for 19.5 days. Edema of calycular neuronal terminals. Destruction of receptor cells, type I. Designations the same as in Figure 2. Electronogram. Magnification 3000.

which is related to the disruption in the formation and efflux of the /26 endo- and perilymph) destruction of individual receptor cells, chiefly of Type I, is observed. Special attention is also drawn to the transformation of the otolith membrane, and in particular, to the transformation of the otoconiae which form in the aggregate the otoliths of the vestibular apparatus. Their loss of their proper crystalline form and the extreme separation of bright and dark substances, which as it were, are detached from each other, indicates some sort of disruption of the calcium exchange within the limits of the otolithic membrane and the otoconiae. We conjecture that the dark substance is an organic ingredient



Figure 4: Otolith membrane (OM) and otoconiae (OT) from the control rat (a); otolith membrane and otoconiae from the rat subjected to the conditions of space flight for 19.5 days (b) Electronogram. Magnification: a-12,000 b- 10,000.

and the bright one, an organic ingredient of the otoconiae. Consequently, the redistribution of these substances suggests disturbances in the calcium metabolism within the otolith and the otolithic membrane. Such a surmise also follows from the observations made by us earlier (Ya. A. Vinnikov, et al., 1976) concerning the otoconiae and the otoliths of amphibians and fishes, and also from the data in the literature concerning the dynamics of the calcium exchange in the otoconiae of the otolithic membrane in mammals in general and in rats in particular (Lim, 1974, 1973; Ross and Peacar, 1975).

Conclusions: (1) With the aid of light and electron microscopes we investigated the vestibular apparatus of 12 rats subjected to weightlessness for 19.5 days on board the satellite "Kosmos-782" and subjected to acceleration during lift-off and touchdown.

(2) In the vestibular apparatus and, in particular, the utricle a number of changes in the type I and II receptor cells and in the otolith membrane were observed which are related to acceleration as well as to the condition of weightlessness.

(3) Acceleration caused the otolithic membrane and the otoliths to be forced against the receptor surface of the utricle and resulted in the peripheral position of the nucleoli inside the nucleus.

(4) 19.5 days of weightlessness was accompanied by increased edema of the tissues of the vestibular apparatus, and may have resulted in destruction of individual receptor cells of type I. Changes in the structure of the otoconiae were also observed which, obviously indicate changes in their calcium ingredient.

(5) In the horizontal canal there was separation of the cupula from the receptor surface of the crista.



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Figure 5: Cupula in the crista of the horizontal canal is pulled off (the rat subjected to the conditions of space flight for 19.5 days). Microphoto. Magnification: oc. 7 ob. 10

Figure 6: The apex of the receptor cell with stereocilia of the crista from the horizontal semicircular canal after the cupula has been pulled off (the rat subjected to the conditions of space flight for 19.5 days). Electronogram. Magnification: 30,000.

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